First report of Plectosphaerella cucumerina on wild rocket (Diplotaxis tenuifolia) in greenhouses in Italy.

First Report of *Plectosphaerella cucumerina* on Wild Rocket (*Diplotaxis tenuifolia*) in Italy. A. Garibaldi, G. Gilardi G. Ortu and M.L. Gullino. AGROINNOVA, Università di Torino Via Leonardo da Vinci 44, 10095 Grugliasco, Italy and

During spring 2012, symptoms of an unusual leaf spot disease were observed in a commercial greenhouses near Salerno (southern Italy) on plants of *Diplotaxis tenuifolia.* The first symptoms on leaves of affected plants consisted of small (1 mm) black-brown spots of irregular shape, later coalescing into larger spots, 1 cm diameter. Spots were surrounded by a yellow halo, and were mostly located on the foliar limb, rib and petiole. Affected leaves often showed distortion, appearing hook-like. The disease was severe under high humidity conditions, at air temperature of 20-26 °C, and caused severe production losses. Particularly, affected tissues rotted quickly after packaging, during transit and commercialization of processed rocket. Diseased tissue was excised, immersed in a solution containing 1% sodium hypochlorite for 60s, rinsed in water, then cultured on potato dextrose agar (PDA) medium, added with 25 mg/l of streptomycin sulphate. After 5 days a fungus developed producing a whitish-orange mycelium when incubated under 12 h/day of fluorescent light at 22 °C. The isolates obtained were purified, subcultured on PDA. On this medium, they produced hyaline elliptical and ovoid conidia, sometimes one-septate, measuring 4.5-9.2 × 1.7-3.5 (average 6.8 x 2,6) µm. Conidia were born on phialides, measuring 6.8-20.2 x1.3-3.1 (average 16.5 x 2.1) µm. Such characteristics are typical of *Plectosphaerella* sp. (2). The ITS region (Internal Transcribed Spacer) of rDNA was amplified using the primers ITS1/ITS4 (3) and sequenced. BLAST analysis of the 519 bp segment showed a 98% similarity with the sequence of *Plectosphaerella cucumerina* (GenBank Accession number AB469880). The nucleotide sequence has been assigned the GenBank Accession JX185769. To confirm pathogenicity, tests were conducted on 45-day-old *D. tenuifolia* plants. Plants (21/treatment), grown in 15 l pots (7 plants/pot) were inoculated by spraying a $1x10^6$ CFU/ml conidial suspension of one isolate of *P. cucumerina,* prepared from 10-day-old cultures, grown on PDA. Inoculated plants were maintained in a growth chamber at 23 ±1°C, at very high relative humidity for 4 days. Non inoculated plants served as control. Inoculated plants showed the typical first leaf spots 6 days after the artificial inoculation. In the following days, spots enlarged and leaves resulted distorted, showing chlorosis. Non-inoculated plants remained healthy. *P. cucumerina* was reisolated from inoculated plants. The pathogenicity test was conducted twice repeating the experimental conditions of the first test and obtaining the same results. This is, to our knowledge, the first report of *P. cucumerina* on *D. tenuifolia* in Italy as well as worldwide. *P. cucumerina* has been described as associated with root and collar rots of other horticultural crops in southern Italy (1). Due to the importance of the crop in Italy, this disease can cause serious economic losses.

References: (1) A. Carlucci *et al*., Persoonia, 28: 34, 2012. (2) M.E. Palm *et al*. Mycologia, 87:397, 1995. (3) T. J. White *et al*. PCR Protocols: A Guide to Methods and Applications. M. A. Innis et al., eds. Academic Press, San Diego, 1990.